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Mutagenesis Screening of Pesticides 'Drosophila'

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MUTAGENESIS SCREENING OF PESTICIDES DROSOPHILA

by

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Contract Number 68-01-2474

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FOREWORD

The many benefits of our modern, developing, industrial society are accompanied by certain hazards. Careful assessment of the relative risk of existing and new man-made environmental hazards is necessary for the establishment of sound regulatory policy. These regulations serve to enhance the quality of our environment in order to promote the public health and welfare and the productive capacity of our Nation's population.

The Health Effects Research Laboratory, Research Triangle Park, conducts a coordinated environmental health research program in toxicology, epidemiology, and clinical studies using human volunteer subjects. These studies address problems in air pollution, non-ionizing radiation, environmental carcinogenesis and the toxicology of pesticides as well as other chemical pollutants. The Laboratory develops and revises air quality criteria documents on pollutants for which national ambient air quality standards exist or are proposed, provides the data for registration of new pesticides or proposed suspension of those already in use, conducts research on hazardous and toxic materials, and is preparing the health basis of non-ionizing radiation standards. Direct support to the regulatory function of the Agency is provided in the form of expert testimony and preparation of affidavits as well as expert advice to the Administrator to assure the adequacy of health care and surveillance of persons having suffered imminent and substantial endangerment of their health.

This report examines the toxicity of 15 selected pesticides and the positive/negative effects upon the Drosophila.

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ABSTRACT

Drosophila melanogaster males were exposed by feeding (plus contact and possibly inhalation). The genetic test found most sensitive and appropriate was the sex-linked recessive lethal test. For this, males of the Canton-S wild type stock were exposed. They were mated individually and brooded to sample the entire range of germ cell developmental stages. Very large number of tests (over 7000) were accumulated for each compound in two or more replicate experiments. Concurrent negative controls were done with each, and positive controls were run occasionally. Thirty pesticides and seven other miscellaneous compounds were tested and four reference mutagens were run through the system, some of the latter at a series of "doses" (exposure concentrations). Table 1 lists them all, with the results.

Of the 15 pesticides (listed first in the table) which could be tested at adequate concentrations, 4 (Captan, Folpet, Bromacil and Simazine) were found to be weak mutagens. One (Cacodylic Acid) was questionable but called negative. The rest of the pesticides were so toxic that only very low concentrations (0.1 - 5 ppm) could be used (usually for a reduced exposure time), and those are not considered adequately tested, in view of results obtained with reference mutagens at these concentrations.

Two of the miscellaneous compounds ("Tris" and $PtCl_4$) were found to be potent mutagens. The rest were negative.

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We also wish to express our appreciation for the assistance of our consultant, Dr. Seymour Abrahamson, Professor of genetics and zoology at the University of Wisconsin.

SUMMARY

Drosophila melanogaster males were exposed by feeding (plus contact and possibly inhalation). The genetic test found most sensitive and appropriate was the sex-linked recessive lethal test. For this, males of the Canton-S wild type stock were exposed. They were mated individually and brooded to sample the entire range of germ cell developmental stages. A very large number of tests (over 7000) were accumulated for each compound in two or more replicate experiments. Concurrent negative controls were done with each, and positive controls were run occasionally. Thirty pesticides and seven other miscellaneous compounds were tested and four reference mutagens were run through the system, some of the latter at a series of "doses" (exposure concentrations). Table 1 lists them all, with the results.

Of the 15 pesticides (listed first in the table) which could be tested at adequate concentrations, four (captan, folpet, bromacil, and simazine) were found to be weak mutagens. One (cacodylic acid) was questionable but called negative. The rest of the pesticides were so toxic that only very low concentrations (0.1-5 ppm) could be used (usually for a reduced exposure time), and those are not considered adequately tested, in view of results obtained with reference mutagens at these concentrations.

Two of the miscellaneous compounds (Tris and $PtCl_4$) were found to be potent mutagens. The rest were negative.

This publication is a summary of the complete project report which can be purchased from the National Technical Information Service.

TABLE 1
SUMMARY OF COMPOUNDS TESTED

Pesticide	Concentration (ppm) in feeding solution	Mutagenesis result
Bromacil	2, 3, 5, 2000	+
Captan	2, 3, 2000	+
Folpet	2, 3, 2000	+
Simazine	3, 5, 2000	+
Cacodylic acid	3, 500, 100	-?
Dicamba	3, 2000	-
DMSA	3, 1500	-
Methoxychlor	1000	-
Monuron	4, 1000, 2000	-
MSMA	2, 3, 4, 1500, 2000	-
Quintozene (PCNB)	3, 4, 8, 2000	-
Trifluralin	2, 3, 1000	-
Siduron	100	-
Acephate	10	-
Carbofuran	10	-
Dimethoate	1, 5, (10)	-
Methomyl	4, (10)	-
Aspon	5	-
Azinphos-methyl (Guthion)	0.25-1.0	-
Chlorpyrifos (Dursban)	0.1	-
Demeton	1	-
Dinoseb	0.5, 1.4	-
Disulfoton	1	-
Ethyl parathion	0.25, 0.5	-
Fenthion	0.1, 0.25	-
Malathion	0.25, 0.5	-
Monocrotophos (Azodrin)	2, 3	-
Phorate (Thimet)	0.5-3	-
Trichlorfon	1	-
Miscellaneous Compounds		
Safrole	100	-
1'Hydroxy Safrole	1000	-
1'Hydroxy Safrole -2,3 epoxide	1000	-
Direct Black 38	2000	-
Tris (2,3-dibromopropyl phosphate)	4, 10, 100, 1000	+
Benzidine dihydrochloride	1000	-
$PtCl_4$	100, 500	+
Reference Mutagens		
EMS (Ethyl methane-sulfonate)	2, 4, 10, 100, 200, 400	+
EI (ethylenimine)	2, 4, 10, 30, 50	+
TMP (trimethyl phosphate)	1000	+
TMP (trimethyl phosphate)	100, 300	-
DBE (1,2-dibromoethane)	5, 10, 50	-?

INTRODUCTION

The mutagenicity tests carried out in this laboratory using Drosophila were part of a larger program involving several test systems in several laboratories. It was not known how Drosophila systems would compare with others in terms of sensitivity to detect mutagenicity. Neither was it known which genetic endpoints (in Drosophila) would be most adequate in a screening test. It was recognized that pesticides (especially the insecticides) would pose special problems for genetic tests with fruit flies. The Drosophila screening system itself was therefore undergoing definition and simplification during the course of these studies.

The original plan was to test each compound in a step-wise manner as follows:

1. Dominant lethal test (the fastest). Stop if positive.
2. Chromosomal alteration test (a one generation test). Stop if positive.
3. Sex-linked recessive lethal test.

It became clear, however, that the first 2 tests were not highly sensitive (thus the lethal test had to be done in all cases) and were not easy to do. At the same time, evidence was mounting (esp. Vogel & Sobels, 1976) that the sex-linked recessive lethal test is by far the most sensitive Drosophila test. It also, of course, includes chromosomal alterations as well as "point" mutations. It was decided, therefore, to run only the sex-linked recessive lethal test. The other genetic schemes were dropped from the program after the first few months and will not be dealt with in detail in this report.

Many people have been hesitant to accept results with an insect as relevant to man. It was thought that insect metabolism was probably too different. These objectives have been considerably reduced by the discovery (Vogel and Sobels, 1976) that Drosophila has enzymes which carry out metabolic activation similar to that effected by mammalian enzyme extracts.

The toxicity of many of the compounds did cause serious problems and resulted in a "no test" conclusion for some of the compounds. For those which could be adequately tested, the Drosophila results compared quite favorably with the other systems, picking up 4 weak mutagens and 1 questionable one. All of these except 1 were picked by at least 1 other system. Two were found positive in all systems.

EXPOSURE METHODS

Initial trials were made of several other exposure methods (aerosol spray, contact, injection) but the method chosen as most appropriate was feeding. Since flies walk on their food, contact is always involved and, when vaporization occurs, the test substance is also inhaled. The method thus seemed more complete and exposure more certain, and it better mimicked human exposure routes.

It was shown several years ago by Lewis and Backer (1968) that feeding of mutagens in glucose solution is effective. They fed flies in vials with a bit of soaked tissue (such as Chem-Wipes). We chose to feed in small disposable petri plates, with a disc of glass fiber filter in the bottom. This permitted easy observation and counting of dead flies for toxicity information.

At first, flies were exposed (toxicity permitting) for 48 hours. Later the time was extended to 72 hours, to make ingestion more certain. Flies cannot survive for more than 24-36 hours without drinking, but in 72 hours they should be obliged to ingest more. Contact (and inhalation) were also thus extended.

Compounds were dissolved, when possible, in 1% glucose. During the first year, DMSO was used for compounds not water soluble. At that time, a decision was taken (after warnings voiced at the Fredericksburg meeting) to avoid DMSO if possible. Thus many compounds were used in suspension rather than in solution.

The concentrations were chosen by the following criteria. If toxic at 2000 ppm or below, a concentration giving as near as possible to 50% mortality at 72 hours was chosen. For very toxic compounds, the duration of exposure was reduced as well as the concentration. For non-toxic compounds, 2000 ppm was chosen as a reasonable upper limit.

SEX-LINKED RECESSIVE LETHAL TEST Introduction

This type of mutation is frequent (involving some 800 genes on the X chromosome) and varied in nature (including both "point" mutations and chromosomal alterations). The test has been found in comparative studies (Vogel and Solels, 1976) to be more sensitive in detecting chemical mutagens thatn other tests in Drosophila. We now feel that the sex-linked recessive lethal test alone should be an adequate probe for mutagenesis.

Stocks and Genetic Scheme

Canton-S wild type males were exposed. "CS" stock was used because it has had a low spontaneous frequency over many years of use. The frequency averages about 0.15% with variations rarely exceeding 0.1% and 0.3%.

Treated males were mated to "FM6" females. The X-chromosome of this is marked with yellow, white and Bar and carries a complex of stock inversions.

Heterozygous F_1 females were mated individually to FM6 males (brothers or stock males) and the F_2 of each was observed for the presence or absence of "+" males. Any culture having no or less than 5% of the expected number of males was considered a lethal case and was confirmed by repeating the test with four individual heterozygous F_2 females mated to FM6 males.

Each treated or control male was transferred to new sets of FM6 females at intervals of 4, 3, 3 and 4 days, to produce 4 broods of

progeny. The different broods sample germ cells in gradually earlier stages of development at the time of exposure. Mature sperm are sampled in brood 1, spermatids in brood 2, spermatocytes in brood 3 and spermatogonia in brood 4. These are not "clean" but rather "rough" samples, due to the length of the mating times and the fact that chemicals may remain in the body beyond the external exposure time.

"Cluster" detection and handling

Treated and control males were numbered, mated and transferred individually and the F_1 daughters of each were mated as a "family." This is done in order to detect cases where more than 1 lethal is produced by 1 male. These cases will be referred to as "multiples" (as opposed to "singles" and "nulls"). A multiple can be due to multiple individual mutations (MIM) or to a single mutation event in a gonial cell, which then reduplicates and produces 2 or more sperm cells carrying the same lethal (a "cluster"). The distinction (if possible) between these two circumstances is very important when attempting to detect low-level mutagenicity. With potent mutagens the contribution of clusters is insignificant and mass matings of treated and control males are the rule. When exposure is simple (as with radiation), and many flies can be exposed, the procedure is often to test only 1 daughter of each treated or control fly, this avoiding both MIMs and clusters. Exposure is not simple with chemicals and thus other precautions must be taken.

Although there are pitfalls which we recognize, we have applied the following procedure to this data. Whenever a multiple was found, a

statistical method (devised by Seymour Abrahamson) was applied, which yields the probability (binomial expectation) of that number of mutations arising independently in a single male given (a) the number of males in that particular treated or control group which produced progency, (b) the average number of F_1 females tested per P_1 male and (c) the mutation frequency.

If the number of lethals in the multiple greatly exceeded probability we considered it a cluster and did not count these lethals.

In addition, however, it was often necessary to make some subjective judgments on particular compounds. When the lethals in the replicate treated groups tended to occur in multiples while those in the concurrent control group did not, this might indicate "spotty" exposure.

The summarized data are presented with presumed clusters removed, but the corrections are indicated at the foot of the table.

Number of chromosomes tested

The number of tests needed per compound or "compound equivalent," which would be any variant on a compound, such as dose level, germ cell stage or exposure method) depends mostly upon (a) mutagenicity - the more mutable, the fewer tests needed to prove it, and (b) the increment in frequency desired to detect.

We have found that when the control rate is 0.15%, and when it is desired to detect (and prove statistically) an equal induced frequency (a doubling of background), then about 8000 treated and 8000 control chromosomes should be tested. Our aim was therefore numbers of this magnitude, accumulated in 2 or 3 replicate experiments.

Results and conclusions

The data from the recessive lethal tests is shown in tables 3 through 52. These are summaries per compound per variation (i.e., exposure concentration or variation in expsoure method). Each is shown with its concurrent controls, which were usually shared with other experiments in the same "runs." Table 3 may be consulted for the cumulative control value (0.157%) and the control frequency variations over time and from run to run. The summary table at the beginning of this report (table 1) shows the concentrations tested and the conclusions as to mutagenicity.

The pesticide results fall into three categories:

(a) Those which were tested at concentrations above 10 ppm for at least 24 hours and which yielded results indicating a weak mutagenic effect. These were Captan, Folpet, Bromacil and Simazine plus possible Cacodylic Acid.

(b) Those which were presumably tested adequately (as above) but which yielded negative results. These were Dicamba through Carbofuran in table 1.

(c) Those which may not have been adequately tested, since they were highly toxic, were used at very low concentrations and gave negative results.

For the four compounds called positive, the conclusion was based upon the experiments with 2000 ppm. Captan, Simazine and Bromacil were tested simultaneously in run no. 37. Folpet was tested in run no. 38. Both runs had controls with quite low frequencies, but this appeared to

be a true low period for the stock. See table 3. Run 37 was done with a newly prepared stock and the control was the first of a series giving low values. In all four cases, there were several multiples in the treated series, but none in the controls. (See page 7 for explanation of multiples.) These multiples were:

Captan - 5 males with 2, 2 males with 3, 1 male with 6

Folpet - 4 males with 2, 1 male with 3

Bromacil - 3 males with 2, 1 male with 3, 1 male with 4, 1 male with 5

Simazine - 4 males with 2, 2 males with 3

When the Kastenbaum and Bowman statistical test (Mut. Res. 9 (1970) 527-549) is applied to the data for these compounds (using the sum of controls 37 and 38 to provide approximately equivalent numbers of treated and control tests), the result is as follows.

Captan is significant at the .01 level if the multiple of 6 is included. Without the multiple, it is significant at the .05 level.

Bromacil is significant at the .01 level if the multiples (one of 4 and one of 5) are included. It barely misses significance at .05 without them.

Simazine had no multiples greater than 3 and is significant at the .01 level.

Folpet also had no ocrrection factor but is significant only t the .05 level. Using the Chi^2 test, which is somewhat less conservative than the Kastenbaum-Bowman test, $P = < .02$. More importantly, the Folpet data shows a peak of mutagenicity in broods 2 and 3. Using these broods only, the result is highly significant ($Chi^2 = 15$, $P = < .01$).

In all these cases, the deduction of multiples of 4, 5, and 6 is questionable, since there are also several multiples of 2 and 3, while the controls have almost none.

It is possible that exposure may have been "spotty" - i.e., some males may have ingested or contacted or inhaled more than others, in which case the compounds may be more mutagenic than the averaged results indicate.

Cacodylic Acid caused marked sterility of treated males, especially in brood 3, indicating a cytotoxic effect on peri-meiotic cells. Unfortunately, it is not possible to know from these experiments whether or not the damage is genetic. The recessive lethal frequency obtained in the one adequate experiment (run 52) was actually 3 times the concurrent control value. The latter, however, was exceptionally low, and in this case it was not part of a consistently low control period. (Controls were in fact somewhat erratic during that time.) There were no multiples in control 52 and only 1 multiple of 3 in the treated males. We thus called the result negative but questionable.

Data from the tests of miscellaneous compounds is shown in tables 37 through 43. Tris and $PtCl_4$ were clearly mutagenic, with peak effects in brood 2, indicating an effect primarily on spermatids. All others were clearly negative.

The reference mutagens EMS, EI and TMP were positive when tested at adequate concentrations. EMS, a very potent mutagen, surprisingly gave frequencies very near control level when tested at very low concentrations (2 and 4 ppm). TMP was negative at 100 ppm and 300 ppm. DBE was tested

only at low concentrations and was negative or questionable. It is probably that air exposure technique was inappropriate for this valatile compound. In all these cases, however, there were multiples, indicating possible spotty exposure. Data for the reference mutagens is in tables 44 through 52.

CHROMOSOME LOSS, REARRANGMENT AND NON-DISJUNCTION

The method consists of treating males whose Y-chromosome has pieces of X-chromosome on both arms, one marked with Y^+ (the normal allele of yellow body) and the other with B^S (Stone's Bar). The F_1 flies are observed for loss of either or both markers. The test is relatively fast, requiring only one generation, and appears to be simple. In use, however, we found that very large numbers are required and interpretation of results was difficult, due to the several different types of variants and their quite different meanings in terms of cytogenetic events. Since the recessive lethal test also picks up chromosomal rearrangements, it was decided that this separate test was not worth the time and effort required.

The data that was collected is shown in table 2. The pesticides gave frequencies varing from 5.31 to 20.29 x 10^{-4}, all with confidence limits overlapping the control frequency. Controls varied from 1.5 to 14.99 x $^{-4}$. Of the three reference mutagens tested, EMS and EI gave clearly positive results, but TMP did not. Evidently it is not an efficient chromosome breaker.

A new scheme, devised by Loring Craymer (1974), came to our attention and was tried in a preliminary manner as a possible replacement for the $Y^+ \cdot Y \cdot B^S$ test. This is a selective scheme, in which the only survivirs are flies resulting from non-disjunction or certain chromosome breakage events. Since no actual tests were performed for this EPA contract, we will not dwell on the test here, but only point out that it is a likely candidate for future screening programs.

DOMINANT LETHALS

Methodology was developed for dominant lethal screening and tests were performed (unfortunately using very low concentrations) on Simazine and Dicamba. Simazine gave possibly positive results. This test, however, was also abandoned, since it was time-consuming and would probably rarely help avoid further testing.

Table 2

Chromosome Rearrangement, Loss and Non-Disjunction

Compounds	Concentration (ppm)	No. Tests	No. Variants	Frequency Variants x 10 -4
Pesticides				
Phorate (Thimet)	1	34,547	32	9.26
Ethyl parathion	0.25, 0.5	20,721	11	5.31
Captan	2, 3	22,387	22	9.83
Folpet	3	16,098	21	13.05
Folpet	2000	5,753	5	8.69
Quintozene (PCNB)	3	4,270	5	11.71
Malathion	0.5	11,701	16	13.67
Bromacil	2, 3	7,125	6	8.42
Azinphos-methyl (Guthion)	0.5	9,792	17	17.36
Monocrotophos (Azodrin)	3	15,030	22	14.64
Fenthion	0.1	13,155	14	10.64
Chlorpyrifos (Dursban)	0.1	12,385	13	10.50
Dinoseb	0.25	3,406	3	8.81
Trifluralin	2, 3	12,013	11	9.16
MSMA	3	3,942	8	20.29
MSMA	1500	6,636	2	3.01
MSMA	2000	938	1	10.66
DSMA	3	8,609	8	9.29
Cacodylic Acid	3	5,937	9	15.16
Dicamba	3	14,396	12	8.34
Simazine	3	15,861	15	9.46
Methomyl	10	20,266	26	12.83
Monuron	4, 8	15,166	15	9.90
Siduron	100	8,409	5	5.90
Acephate	10	9,073	8	8.82
Methoxychlor	1000	9,971	6	6.02
Reference Mutagens				
EMS	400	1,561	27	172.97
EI	50	1,302	20	153.61
TMP	1000	7,515	12	15.97
All Controls	-	101,520	80	7.88

Table 3

Summary of controls concurrent with pesticide runs or interspersed with them. In column headed "No. lethals", S=single, M = multiple, T = total. See explanation of multiples, p.7.

Table 3

CONTROLS

Run	No. tests (1/1+) per brood 1	2	3	4	Total Tests	No. lethals S	M	T	% lethals
1-6	14/12048	-	-	-	12,062	14	0	14	0.116
7-11	7/5682	-	-	-	5689	5	1(2)	7	0.123
12-14	5/1538	-	-	-	1543	3	1(2)	5	0.324
17	0/783	0/104	1/317	1/178	1384	2	0	2	0.145
18	3/1249	2/543	0/270	1/664	2732	2	2(2)	6	0.220
20	4/2291	0/557	0/150	2/717	3721	6	0	6	0.161
22	0/711	2/584	0/273	1/426	1997	3	0	3	0.150
24	1/1291	2/630	2/783	0/786	3495	5	0	5	0.143
25	0/384	-	-	-	384	0	0	0	0
26	4/1738	0/349	1/613	0/594	3299	2	1(3)	5	0.152
27	2/1272	2/661	0/290	0/610	2837	4	0	4	0.141
28(obs.)	9/1322	1/362	3/741	8/770	3216	5	1(3) 1(6) 1(7)	21	0.653
28(corr.)	6/1322	0/362	2/741	0/770	3203	5	1(3)	8	0.249
31(obs.)	4/1739	2/1091	18/681	10/748	4294	5	1(2) 1(28)	35	0.815
31(corr.)	3/1739	0/1091	3/681	1/748	4266	5	1(2)	7	0.164
32	-	2/552	0/341	3/1091	1989	3	1(2)	5	0.251
33(obs.)	4/1295	10/1302	10/1355	0/728	4704	3	3(2) 1(3) 1(12)	24	0.510
33(corr.)	4/1295	4/1302	4/1355	0/728	4692	3	3(2) 1(3)	12	0.255
35	0/609	2/415	1/486	2/564	2079	5	0	5	0.240
36	6/1591	0/419	-	-	2016	2	2(2)	6	0.298
37	4/1573	1/1438	1/1087	0/650	4754	6	0	6	0.126
38	1/1720	3/1102	0/803	0/1187	4816	4	0	4	0.083
39	3/1567	0/762	1/700	0/654	3687	2	1(2)	4	0.108
40	3/2792	0/793	1/611	1/814	5015	5	0	5	0.100
41	0/2449	4/1413	3/1843	1/1076	6789	5	1(3)	8	0.118
42	3/2882	2/1902	0/1197	2/1148	7136	7	0	7	0.098
43(obs.)	11/2081	10/1708	8/1100	2/1052	5972	7	2(2) 2(3) 1(14)	31	0.519

(continued)

CONTROLS (CONTINUED)

Run	No. tests (1/1+) per brood 1	2	3	4	Total Tests	No. lethals S	M	T	% lethals
43(corr.)	3/2081	5/1708	7/1100	2/1052	5958	7	2(2) 2(3)	17	0.285
44	8/2622	3/1359	1/1236	1/767	5997	8	1(2) 1(3)	13	0.217
45	2/1636	0/635	5/1355	3/507	4143	3	2(2) 1(3)	10	0.241
46	9/1120	0/605	2/926	0/701	3363	8	1(3)	11	0.327
47	2/1946	0/912	1/910	0/510	4281	3	0	3	0.070
48	7/1429	5/1078	2/842	3/464	3830	9	4(2)	17	0.444
49	3/1310	2/1357	3/1116	1/700	4492	9	0	9	0.200
50	3/1705	2/1250	3/1336	3/1170	5472	5	3(2)	11	0.201
51	8/1895	1/1192	1/1076	1/541	4715	9	1(2)	11	0.233
52	0/1461	2/2337	1/1727	0/814	6342	3	0	3	0.047
53	3/1809	-	2/1447	0/912	4173	5	0	5	0.120
54	0/1934	0/856	2/1284	0/606	4682	2	0	2	0.043
55	6/1480	1/435	0/853	1/409	3185	6	1(2)	8	0.251
56	1/991	1/1129	0/1265	1/541	3929	3	0	3	0.076
57	1/1474	1/1502	1/1337	0/679	4995	3	0	3	0.060
58(obs.)	1/1264	1/936	10/1464	0/927	4603	3	1(2) 1(3) 1(4)	12	0.261
58(corr.)	1/1264	1/936	6/1464	0/927	4599	3	1(2) 1(3)	8	0.174
59	2/2086	4/1681	1/974	1/923	5672	8	0	8	0.141
60	1/1345	0/738	3/1427	1/909	4424	5	0	5	0.113
61(obs.)	0/1166	0/702	4/887	4/546	3309	3	1(5)	8	0.242
61(corr.)	0/1166	0/702	2/887	1/546	3304	3	0	3	0.091
62	0/2269	0/677	1/966	1/820	4734	2	0	2	0.042
1-62(obs.)	146/81549	68/36068	93/36069	55/27903	181951	202	27(2) 10(3) 1(4) 1(5) 1(6) 1(7) 1(12) 1(14) 1(28)	362	0.199
% lethals per brood	0.179	0.188	0.257	0.197					
1-62(corr.)	133/81549	54/36068	64/36069	35/27903	181875	202	27(2) 10(3)	286	0.157
% lethals per brood	0.163	0.149	0.177	0.125					

Tables 4 through 36

Summary of recessive lethal experiments per pesticide, with the concurrent controls. Compounds in alphabetical order. Conc. (ppm) = the concentration of the compound in the 1% glucose exposure solution. Time (hrs.) = the duration of exposure. % Mort. = the percent of exposed flies who died before the end of the exposure. When appreciable death occurred after mating, this is noted in a footnote. Run = a group of experiments sharing one control and started on a particular date. These "starts" were numbered consecutively. Whenever presumed "clusters" were corrected for, the correction is shown in a footnote.

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Table 4

ACEPHATE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	10	48	38	40	4/2443	1/683	2/207	3/1246	4589	10	0.218
C	10	48	83	57	0/1666	0/1490	2/1167	1/668	4994	3	0.060
Sum experiments					4/4109	1/2173	4/1374	4/1914	9583	13	0.136
% lethals per brood					0.100	0.046	0.290	0.290			
Controls (concurrent)											
			1	40	3/2792	0/793	1/611	1/814	5015	5	0.099
			6	57	1/1474	1/1502	1/1337	0/679	4995	3	0.06
Sum controls					4/4266	1/2295	2/1948	1/1493	10,010	8	0.079
% lethals per brood					0.094	0.044	0.103	0.067			

Table 5

ASPON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^{+}$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	5	8	90	41	0/358	0/266	0/263	0/238	1125	0	0
B	5	2-2-4	12	42	0/2553	3/2031	3/1288	1/974	6853	7	0.102
Sum experiments					0/2911	3/2297	3/1551	1/1212	7978	7	0.088
% lethals per brood					0	0.130	0.193	0.083			
Controls (concurrent)											
			2	41	0/2449	4/1413	3/1843	1/1076	6789	8	0.117
			3	42	3/2882	2/1902	0/1197	2/1148	7136	7	0.098
Sum controls					3/5331	6/3315	3/3040	3/2224	13,925	15	0.107
% lethals per brood					0.056	0.181	0.099	0.135			

Table 6
AZINPHOS-METHYL

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	0.25	48	36	17	0/1201	1/748	1/398	1/347	2697	3	0.111
	0.50	48	44	17							
	1.0	48	56	17							
C	0.5	48	42	27	0/1561	0/728	0/453	4/670	3416	4	0.117
Sum experiments					0/2762	1/1476	1/851	5/1017	6113	7	0.114
% lethals per brood					0.0	0.067	0.117	0.491			
Controls (concurrent)											
			52	17	0/783	0/104	1/317	1/178	1384	2	0.144
			5	27	2/1272	2/661	0/290	0/610	2837	4	0.140
Sum controls					2/2055	2/765	1/607	1/788	4221	6	0.142
% lethals per brood					0.097	0.261	0.164	0.127			

Table 7

BROMACIL

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A-C	2,3	48	24-64	12-14	17/5551	-	-	-	5568	17	0.305
D	3	48	18	22	1/647	0/641	0/270	2/628	2189	3	0.137
Sum experiments					18/6198	0/641	0/270	2/628	7757	20	0.258
% lethals per brood					0.290	0	0	0.318			
Controls (concurrent)											
			0-54	12-14	5/1538	-	-	-	1543	5	0.324
			0	22	0/711	2/584	0/273	1/426	1997	3	0.15
Sum controls					5/2249	2/584	0/273	1/426	3540	8	0.226
% lethals per brood					0.222	0.341	0	0.234			

Table 8

BROMACIL

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
G	2000	48	2	37	6/3163	5/2186	3/1824	3/1327	8517	17*	0.199
I	2000	72	24	54	-	-	1/1595	-	1596	1	0.063
Sum experiments					6/3163	5/2186	4/3419	3/1327	10,113	18	0.178
% lethals per brood					0.189	0.228	0.117	0.226			
Controls (concurrent)											
			1	37	4/1573	1/1438	1/1087	0/650	4754	6	0.126
			3	54	0/1934	0/856	2/1284	0/606	4682	2	0.042
Sum controls					4/3507	1/2294	3/2371	0/1256	9436	8	0.084
% lethals per brood					0.114	0.044	0.126	0			

*Cluster of 4 and cluster of 5 deducted

Table 9

BROMACIL*

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
J	5	72(A)	9(A)	56	3/632	2/851	0/1126	1/307	2922	6	0.205
L	5	72(A)	50(A)	58	2/1217	0/1241	4/1668	1/1173	5306	7	0.132
J,L					5/1849	2/2092	4/2794	2/1480	8228	13	0.158
K	5	-	-	56	0/615	1/886	0/831	0/399	2732	1	0.037
M	5	-	-	58	0/1409	0/1353	1/1848	1/1313	5929	6	0.101
K,M					0/2024	1/2239	1/2679	1/1712	8661	7	0.081
Sum experiments					5/3873	3/4331	5/5473	3/3192	16,889	20	0.118
% lethals per brood					0.129	0.069	0.091	0.094			
Controls (concurrent)											
			2	56	1/991	1/1129	0/1265	1/541	3929	3	0.076
			0	58	1/1264	1/936	6/1464	0/927	4599	8**	0.173
Sum controls					2/2255	2/2065	6/2729	1/1468	8528	11	0.128
% lethals per brood					0.089	0.097	0.219	0.068			

* J,L: Larvae and adults exposed, K,M: Larvae only exposed
** Cluster of 4 deducted

Table 10

CACODYLIC ACID

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
B	3	48	16	24	2/2032	5/1978	0/1394	1/1607	7,019	8	0.113
D	1000	48	55	48	0/789	0/124	0/101	0/24	1,038	0	0.0
E	1000	48	60	50	3/1078	0/61	0/55	0/2	1,199	3	0.250
F	500	72	61	52	6/6036	4/2159	0/22	0/4	8,231	10	0.121
Sum experiments					11/9935	9/4322	0/1572	1/1637	17,487	21	0.120
% lethals per brood					0.110	0.208	0.0	0.061			
Controls (concurrent)											
			-	24	1/1291	2/630	2/783	0/786	3,495	5	0.143
			6	48	7/1429	5/1078	2/842	3/464	3,830	17	0.443
			18	50	3/1705	2/1250	3/1336	3/1170	5,472	11	0.201
			12	52	0/1461	2/2337	1/1727	0/814	6,342	3	0.047
Sum controls					11/5886	11/5295	8/4688	6/3234	19,139	36	0.188
% lethals per brood					0.187	0.207	0.170	0.185			

Table 11

CAPTAN

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2,3	48	10,32	17	0/1024	0/971	0/388	2/395	2782	4*	0.144
B	3	48	32	27	4/1051	3/623	0/296	1/416	2395	8	0.334
D	3	48	4	32	1/380	4/1532	2/1025	3/2314	5261	10	0.190
E	2000	48	2	37	7/2866	7/2607	5/2103	2/1333	8930	21**	0.235
Sum experiments					12/5321	16/5733	7/3812	8/4458	19,368	43	0.222
% lethals per brood					0.225	0.278	0.183	0.179			
Controls (concurrent)											
			52	17	0/783	0/104	1/317	1/178	1384	2	0.145
			5	27	2/1272	2/661	0/290	0/610	2837	4	0.141
			0	32	-	2/552	0/341	3/1091	1989	5	0.251
			1	37	4/1573	1/1438	1/1087	0/650	4754	6	0.126
Sum controls					6/3628	5/2755	2/2035	4/2529	10,964	17	0.155
% lethals per brood					0.165	0.131	0.098	0.158			

* Cluster of 5 deducted
** Cluster of 6 deducted

Table 12

CARBOFURAN

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	10	24	55	56	2/1147	1/1179	3/1551	0/579	4462	6	0.134
B	10	24	89	57	2/2219	0/1924	0/964	1/834	5944	3	0.050
Sum experiments					4/3366	1/3103	3/2515	1/1413	10,406	9	0.086
% lethals per brood					0.118	0.032	0.119	0.070			
Controls (concurrent)											
			2	56	1/991	1/1129	0/1265	1/541	3929	3	0.076
			6	57	1/1474	1/1502	1/1337	0/679	4995	3	0.060
Sum controls					2/2465	2/2631	1/2602	1/1220	8924	6	0.067
% lethals per brood					0.081	0.076	0.038	0.082			

Table 13

CHLORPYRIFOS

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	0.1	48	32	18	3/1524	3/1176	2/931	1/1067	4707	9*	0.191
B	0.1	48	2	26	1/1346	1/730	0/474	0/384	2936	2	0.068
Sum experiments					4/2870	4/1906	2/1405	1/1451	7643	11	0.143
% lethals per brood					0.139	0.209	0.142	0.068			
Controls (concurrent)											
			-	18	3/1249	2/543	0/270	1/664	2732	6	0.219
			4	26	4/1738	0/349	1/613	0/594	3299	5	0.151
Sum controls					7/2987	2/892	1/883	1/1258	6031	11	0.182
% lethals per brood					0.234	0.224	0.113	0.079			

* Cluster of 10 deducted

Table 14

DEMETON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1	2	46	42	1/2255	2/1433	1/1338	1/836	5867	5	0.085
B	1	72	82	51	4/991	1/794	0/458	0/350	2598	5	0.192
Sum experiments					5/3246	3/2227	1/1796	1/1186	8465	10	0.118
% lethals per brood					0.154	0.134	0.055	0.084			
Controls (concurrent)											
			3	42	3/2882	2/1902	0/1197	2/1148	7136	7	0.098
			12	51	8/1895	1/1192	1/1076	1/541	4715	11	0.233
Sum controls					11/4777	3/3094	1/2273	3/1689	11,851	18	0.151
% lethals per brood					0.230	0.097	0.044	0.177			

Table 15

DICAMBA

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
B	3	48	42	25	0/256	-	-	-	256	0	0.0
C	3	48	0	28	9/2778	5/1002	2/1409	1/2204	7410	17	0.229
D	2000	72	16	48	3/1886	2/1110	3/883	0/644	4531	8	0.176
Sum experiments					12/4920	7/2112	5/2292	1/2848	12,197	25	0.204
% lethals per brood					0.243	0.331	0.218	0.035			
Controls (concurrent)											
			4	25	0/384	-	-	-	384	0	0.0
			0	28	6/1322	0/362	2/741	0/770	3203	8*	0.249
			6	48	7/1429	5/1078	2/842	3/464	3830	17	0.443
Sum controls					13/3135	5/1440	4/1583	3/1234	7417	25	0.337
% lethals per brood					0.413	0.346	0.252	0.243			

* Cluster of 6 and cluster of 7 deducted

Table 16

DINOSEB

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
B	0.5,1	48	28,22	20	4/2007	1/709	0/150	2/786	3659	7	0.191
C	4	48	6	35	1/790	0/1153	4/1072	8/1458	4486	13	0.289
Sum experiments					5/2797	1/1862	4/1222	10/2244	8145	20	0.245
% lethals per brood					0.178	0.053	0.327	0.445			
Controls (concurrent)											
			-	20	4/2291	0/557	0/150	2/717	3721	6	0.161
			3	35	0/609	2/415	1/486	2/564	2079	5	0.240
Sum controls					4/2900	2/972	1/636	4/1281	5800	11	0.189
% lethals per brood					0.138	0.205	0.157	0.311			

Table 17

DIMETHOATE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^{+}$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	5	5	40	42	4/2840	1/1915	2/2076	1/1285	3124	8	0.098
B,C	1,10	72	20,28	51	3/1745	0/1914	0/1097	1/750	5510	4	0.072
Sum experiments					7/4585	1/3829	2/3173	2/2035	13,634	12	0.088
% lethals per brood					0.152	0.026	0.063	0.098			
Controls (concurrent)											
			3	42	3/2882	2/1902	0/1197	2/1148	7136	7	0.098
			12	51	7/1895	1/1192	1/1076	1/541	4715	11	0.233
Sum controls					10/4777	3/3094	1/2273	3/1689	11,851	18	0.151
% lethals per brood					0.209	0.097	0.044	0.177			

Table 18

DISULFOTON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1	4	16	61	0/463	0/351	0/306	0/182	1302	0	0.0
B	1	5	18*	62	3/3146	1/919	1/1587	2/1094	6753	7	0.103
Sum experiments					3/3609	1/1270	1/1893	2/1276	8055	7	0.086
% lethals per brood					0.083	0.078	0.052	0.156			
Controls (concurrent)											
			0	61	0/1166	0/702	2/887	1/546	3304	3**	0.09
			0	62	0/2269	0/677	1/966	1/820	4734	2	0.042
Sum controls					0/3435	0/1379	3/1853	2/1366	8038	5	0.062
% lethals per brood					0	0	0.162	0.146			

* 23% more died soon after mating
** Cluster of 5 deducted

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Table 19

DSMA

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	3	48	32	24	4/2240	1/1537	2/1666	1/1378	6829	8	0.117
B	1500	48	56	45	2/1361	1/798	0/1079	0/585	3826	3*	0.078
Sum experiments					6/3601	2/2335	2/2745	1/1963	10,655	11	0.103
% lethals per brood					0.166	0.085	0.072	0.050			
Controls (concurrent)											
			-	24	1/1291	2/630	2/783	0/786	3595	5	0.143
			2	45	2/1636	0/635	5/1355	3/507	4143	10	0.241
Sum controls					3/2927	2/1265	7/2138	3/1293	7638	15	0.196
% lethals per brood					0.102	0.158	0.326	0.231			

* Cluster of 16 deducted

Table 20

FENSULFOTHION

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1	24	0	56	1/1321	3/1504	2/1664	0/681	5177	7	0.135
B	1	48	20*	58	2/1368	2/1202	4/1673	4/1360	5615	12	0.213
Sum experiments					3/2689	5/2706	6/3337	4/2041	10,792	19	0.176
% lethals per brood					0.111	0.184	0.179	0.195			
Controls (concurrent)											
			2	56	1/991	1/1129	0/1265	1/541	3929	3	0.076
			0	58	1/1264	1/936	6/1464	0/927	4599	8**	0.173
Sum controls					2/2255	2/2065	6/2729	1/1468	8528	11	0.128
% lethals per brood					0.089	0.097	0.219	0.068			

* 20% mortality at 48 hrs. (time of mating), 27.5% of those mated died soon and 1 of 29 remaining was sterile
** Cluster of 4 deducted

Table 21

FENTHION

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	0.1,0.25	48,48	4,20	18	6/1703	4/1312	1/1051	0/1056	5122	11	0.214
C	0.1	48	4	26	2/1222	1/638	0/325	0/241	2429	3	0.123
Sum experiments					8/2925	5/1950	1/1376	0/1297	7562	14	0.185
% lethals per brood					0.273	0.256	0.072	0.0			
Controls (concurrent)											
			-	18	3/1249	2/543	0/270	1/664	2732	6	0.219
			4	26	4/1738	0/349	1/613	0/594	3299	5	0.151
Sum controls					7/2987	2/892	1/883	1/1258	6031	11	0.182
% lethals per brood					0.234	0.224	0.113	0.079			

Table 22

FOLPET

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2,3	48,48	18,46	20	1/1876	1/587	0/443	1/438	3347	3	0.090
C	3	48	0	32	1/422	1/871	1/1302	5/1670	4273	8*	0.187
D	2000	72	0	38	4/2857	7/1360	6/1449	3/1719	7405	20	0.270
Sum experiments					6/5155	9/2818	7/3194	9/3827	15,025	31	0.206
% lethals per brood					0.116	0.319	0.219	0.235			
Controls (concurrent)											
			-	20	4/2291	0/557	0/150	2/717	3721	6	0.161
			0	32	-	2/552	0/341	3/1091	1989	5	0.251
			4	38	1/1720	3/1102	0/803	0/1187	4816	4	0.083
Sum controls					5/4011	5/2211	0/1294	5/2995	10,526	15	0.142
% lethals per brood					0.125	0.226	0	0.167			

* Cluster of 4 and cluster of 11 deducted

Table 23

MALATHION

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	0.25 } 0.5	48	8	17	2/1040	3/866	1/399	0/471	2782	6*	0.214
C	0.5	48	40	27	2/1553	1/918	0/555	1/800	3830	4	0.104
Sum experiments					4/2593	4/1784	1/954	1/1271	6612	10	0.151
% lethals per brood					0.154	0.224	0.104	0.078			
Controls (concurrent)											
			52	17	0/783	0/104	1/317	1/178	1384	2	0.144
			5	27	2/1272	2/661	0/290	0/610	2837	4	0.140
Sum controls					2/2055	2/765	1/607	1/788	4221	6	0.142
% lethals per brood					0.097	0.261	0.164	0.127			

* Cluster of 21 deducted

Table 24

METHOMYL

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^{+}$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
C^1	10	48	(high)	31	0/543	1/415	2/308	0/149	1418	3	0.211
C^2	4	48	78	31	3/1762	1/1701	2/1408	1/955	5833	7	0.120
Sum experiments					3/2305	2/2116	4/1716	1/1104	7251	10	0.137
Controls (concurrent)											
			0	31	3/1739	0/1091	3/681	1/748	4266	7*	0.164
% lethals per brood					0.172	0	0.439	0.134			

* Cluster of 28 deducted

Table 25

METHOXYCHLOR

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1000	72	7	40	3/2501	1/736	0/734	0/714	4689	4	0.085
B	1000	72	18	41	2/1943	2/841	3/938	3/859	4591	10*	0.217
Sum experiments					5/444	3/1577	3/1672	3/1573	9280	14	0.150
% lethals per brood					0.112	0.19	0.179	0.19			
Controls (concurrent)											
			1	40	3/2792	0/793	1/611	1/814	5015	5	0.099
			2	41	0/2449	4/1413	3/1843	1/1076	6789	8	0.117
Sum controls					3/5241	4/2206	4/2454	2/1890	11,804	13	0.110
% lethals per brood					0.057	0.181	0.163	0.106			

* Cluster of 17 deducted

Table 26

MONOCROTOPHOS

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^{+}$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2	48	4	18	4/1753	0/1267	0/831	0/943	4794	4	0.083
C	3	48	4	26	3/1368	2.852	3/734	2/518	3482	10	0.287
Sum experiments					7/3121	2/2119	3/1565	2/1461	8276	14	0.169
% lethals per brood					0.224	0.094	0.191	0.137			
Controls (concurrent)											
			-	18	3/1249	2/543	0/270	1/664	2732	6	0.219
			4	26	4/1738	0/349	1/613	0/594	3299	5	0.151
Sum controls					7/2987	2/892	1/883	1/1258	6031	11	0.182
% lethals per brood					0.234	0.224	0.113	0.079			

Table 27

MONURON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
C	4	48	4	31	5/3055	0/1639	4/852	0/1610	7165	9*	0.125
D	1000	72	66	48	1/1556	6/1051	1/573	0/469	3657	8	0.218
	2000	72	68(1)								
Sum experiments					6/4611	6/2690	5/1425	0/2079	10,822	17	0.157
% lethals per brood					0.13	0.223	0.35	0.0			
Controls (concurrent)											
			0	31	3/1739	0/1091	3/681	1/748	4266	7**	0.164
			6	48	7/1429	5/1078	2/842	3/464	3830	17	0.443
Sum controls					10/3168	5/2169	5/1523	4/1212	8096	24	0.296
% lethals per brood					0.315	0.230	0.327	0.329			

* Cluster of 4 deducted
** Cluster of 28 deducted
(1) Most died soon after mating

Table 28

NSMA

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2,3	48,48	18,14	20	2/2224	0/409	0/57	0/728	3420	2	0.058
D	4	48	7	35	2/602	1/829	2/1099	1/726	3262	6	0.183
E	2000	48	91	38	1/498	2/99	0/145	0/125	870	3	0.344
F	1500	24	47	39	5/1775	1/821	1/1023	1/812	4439	8	0.180
Sum experiments					10/5099	4/2158	3/2324	2/2391	11,991	19	0.158
% lethals per brood					0.196	0.185	0.129	0.084			
Controls (concurrent)											
			-	20	4/2291	0/557	0/150	2/717	3721	6	0.161
			3	35	0/609	2/415	1/486	2/564	2079	5	0.240
			4	38	1/1720	3/1102	0/803	0/1187	4816	4	0.083
			0	39	3/1567	0/762	1/700	0/654	3687	4	0.108
Sum controls					8/6187	5/2836	2/2139	4/3122	14,303	19	0.132
% lethals per brood					0.129	0.176	0.093	0.128			

Table 29

PARATHION

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A-E	0.25,0.5	48	0-72	7-11	16/11,914	-	-	-	11,930	16	0.134
Controls (concurrent)			0-8	7-11	7/5682	-	-	-	5689	7	0.123

Table 30

PCNB

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	8	48	20	17	0/751	1/553	1/254	0/233	1793	2	0.111
B	3	48	10	22	2/726	1/536	1/264	2/621	2153	6	0.278
C	4	48	2	35	1/749	1/788	3/1347	3/789	3681	8	0.217
D	2000	24	-	39	6/1587	3/1524	2/1706	1/1339	6168	12	0.194
Sum experiments					9/3813	6/3401	7/3571	6/2982	13,795	28	0.202
% lethals per brood					0.236	0.176	0.196	0.201			
Controls (concurrent)											
			52	17	0/783	0/104	1/317	1/178	1384	2	0.144
			0	22	0/711	2/584	0/273	1/426	1997	3	0.150
			3	35	0/609	2/415	1/486	2/564	2079	5	0.240
			0	39	3/1567	0/762	1/700	0/654	3687	4	0.108
Sum controls					3/3670	4/1865	3/1776	4/1822	9147	14	0.153
% lethals per brood					0.082	0.214	0.169	0.219			

Table 31

PHORATE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A-F	0.5	48	6								
	1	48	4-94								
	2	24,48	28,66	1-6	23,15,450	-	-	-	15,473	23	0.148
	3	48	88								
Controls (concurrent)			0-6	1-6	14/12,048	-	-	-	12,062	14	0.116

Table 32

SIDURON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	100	72	0	40	5/2198	0/604	1/466	0/1020	4294	6	0.139
B	100	72	2	41	1/2487	0/1022	1/1203	2/815	5530	3*	0.054
C	100	72	13	50	1/444	2/1092	3/1268	1/912	4723	7	0.148
D	100	72	6	50	2/1363	3/1142	2/1090	0/661	4263	7	0.164
Sum experiments					9/7492	5/3860	6/4027	3/3408	18,810	23	0.122
% lethals per brood					0.120	0.129	0.148	0.088			
Controls (concurrent)											
			1	40	3/2792	0/793	1/611	1/814	5015	5	0.099
			2	41	0/2449	4/1413	3/1843	1/1076	6789	8	0.117
			18	50	3/1705	2/1250	3/1336	3/1170	5472	11	0.201
Sum controls					6/6946	6/3456	7/3790	5/3060	17,276	24	0.138
% lethals per brood					0.086	0.173	0.184	0.163			

* Cluster of 4 deducted

Table 33

SIMAZINE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^{+}$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
B	3	48	4	25	7/2029	-	-	-	2036	0*	0
C	3	48	11	28	9/1893	5/999	4/1868	0/1680	6458	18	0.279
E	2000	48	3	37	10/3106	8/2212	4/1752	3/1133	8228	25	0.304
Sum experiments					26/7028	13/3211	8/3620	3/2813	16,722	43	0.257
% lethals per brood					0.369	0.403	0.221	0.107			
Controls (concurrent)											
			4	25	0/384	-	-	-	384	0	0
			0	28	6/1322	0/362	2/741	0/770	3203	8**	0.249
			1	37	4/1573	1/1438	1/1087	0/650	4754	6	0.126
Sum controls					10/3279	1/1800	3/1828	0/1420	8341	14	0.168
% lethals per brood					0.304	0.056	0.164	0			

* Cluster of 7 deducted
** Cluster of 6 and cluster of 7 deducted

Table 34

SIMAZINE " CHRONIC" EXPOSURE

G: Larve and adults exposed
H: Larve only exposed

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
G	5	72(A)	2(A)	60	4/1674	3/758	3/1443	0/999	4884	10*	0.204
H	5	-	-	60	4/1647	0/788	0/1357	0/940	4736	4	0.084
Sum experiments					8/3321	3/1546	3/2800	0/1939	9620	14	0.146
% lethals per brood					0.240	0.194	0.107	0			
Controls (concurrent)											
			0	60	1/1345	0/738	3/1427	1/909	4424	5	0.113
% lethals per brood					0.074	0	0.210	0.110			

* Cluster of 18 deducted

Table 35

TRICHLORFON

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1	2	0	61	1/861	0/737	1/948	1/653	3202	3	0.093
B	1	3	12[1]	62	4/2436	2/1059	1/1418	6/1514	6440	13	0.201
Sum experiments					5/3297	2/1796	2/2366	7/2167	9642	16	0.165
% lethals per brood					0.151	0.111	0.084	0.323			
Controls (concurrent)											
			0	61	0/1166	0/702	2/887	1/546	3304	3*	0.09
			0	62	0/2269	0/677	1/966	1/820	4734	2	0.042
Sum controls					0/3435	0/1379	3/1853	2/1366	8038	5	0.062
% lethals per brood					0	0	0.162	0.146			

* Cluster of 5 deducted

(1) 12% at 3 hours, 30% next day at mating time, plus 7 of 50 mated died soon after and 2 were sterile.

Table 36

TRIFLURALIN

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2,3	48,48	20,18	18	2/1637	3/865	0/803	2/1015	4327	7*	0.161
B	3	48	4	26	1/1190	3/666	1/508	1/407	2777	6	0.216
C	1000	72	12	45	1/1523	3/783	4/1527	3/806	4650	11	0.236
D	1000	72	72	50	5/2194	1/1761	6/1713	1/1450	7151	13	0.181
Sum experiments					9/6544	10/4095	11/4551	7/3678	18,905	37	0.195
% lethals per brood					0.137	0.244	0.241	0.190			
Controls (concurrent)											
			-	18	3/1249	2/543	0/270	1/664	2732	6	0.219
			4	26	4/1738	0/349	1/613	0/594	3299	5	0.151
			2	45	2/1636	0/635	5/1355	3/507	4143	10	0.241
			18	50	3/1705	2/1250	3/1366	3/1170	5472	11	0.201
Sum controls					12/6328	4/2777	9/3574	7/2935	15,646	32	0.204
% lethals per brood					0.189	0.144	0.215	0.238			

* Cluster of 5 deducted

Tables 37 through 43

Summary of recessive lethal experiments testing compounds other than pesticides.

Table 37

BENZIDINE DIHYDROCHLORIDE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1000	72	50	63	3/2015	2/1062	2/578	1/885	4548	8*	0.176
B	1000	72	32	64	2/1610	1/944	1/1147	1/635	4341	5	0.115
Sum experiments					5/3625	3/2006	3/1725	2/1520	8889	13	0.146
% lethals per brood					0.138	0.149	0.174	0.131			
Controls (concurrent)											
			4	63	3/2195	1/1097	1/753	2/825	4877	7	0.144
			1	64	3/1535	3/1124	1/808	1/572	4047	8	0.198
Sum controls					6/3730	4/2221	2/1561	3/1397	8924	15	0.168
% lethals per brood					0.061	0.180	0.128	0.214			

* Cluster of 4 deducted

Table 38

CARCINOGENS*

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	100	72	10	59	2/2262	2/2134	2/1040	1/994	6437	7	0.109
B	1000	72	69	59	0/2358	3/1802	1/406	1/770	5431	5	0.092
C	1000	72	4	59	3/2293	3/1748	3/650	2/870	5572	11	0.197
Controls (concurrent)											
			4	59	2/2086	4/1681	1/974	1/923	5672	8	0.141

* A: Safrole B: 1' Hydroxy Safrole C: 1' Hydroxy Safrole - 2, 3 epoxide

Table 39

DIRECT BLACK 38

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	2000	72	4	63	0/2054	1/887	3/1158	0/880	4983	4	0.080
B	2000	72	2	64	1/1574	3/1165	0/1034	1/630	4408	5*	0.113
Sum experiments					1/3628	4/2052	3/2192	1/1510	9400	9	0.096
% lethals per brood					0.028	0.195	0.137	0.066			
Controls (concurrent)											
			4	63	3/2195	1/1097	1/753	2/825	4877	7	0.144
			1	64	3/1535	3/1124	1/808	1/572	4047	8	0.198
Sum controls					6/3730	4/2221	2/1561	3/1397	8924	15	0.168
% lethals per brood					0.161	0.180	0.128	0.214			

* Cluster of 9 deducted

Table 40

$PTCL_4$

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	100	72	36	65	3/1030	20/1060	4/746	2/756	3721	29	0.779
C	100	72	73	66	3/1085	10/528	2/119	2/445	2194	17	0.775
A,C					6/2115	30/1588	6/865	4/1201	5915	46	0.778
B	500	48	60	65	1/952	13/1125	12/760	0/547	3410	26	0.762
D	500	48	55	66	6/943	10/690	2/527	2/704	2884	20	0.693
B,D					7/1895	23/1815	14/1287	2/1251	6294	46	0.731
Sum experiments					13/4010	53/3403	20/2152	6/2452	12,209	92	0.754
% lethals per brood					0.323	1.53	0.921	0.244			
Controls (concurrent)											
			3	65	0/1160	0/961	2/1256	0/617	3994	2	0.050
			3	66	0/998	1/862	0/452	0/407	2709	1	0.037
Sum controls					0/2148	1/1823	2/1708	0/1024	6703	3	0.045
% lethals per brood					0	0.055	0.117	0			

Table 41

"TRIS"

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
C	4	72	15	68	4/1241	0/543	0/637	0/859	3284	4	0.121
F	4	72	24	69	4/1332	2/1109	2/457	0/689	3595	8	0.223
C,F					8/2573	2/1652	2/1094	0/1548	6879	12	0.174
D	10	72	21	68	2/1224	2/821	5/552	1/807	3414	10	0.292
G	10	72	31	69	2/1565	1/1125	0/616	0/844	4153	3	0.072
D,G					4/2789	3/1946	5/1168	1/1651	7567	13	0.172
Sum of experiments					12/5362	5/3598	7/2262	1/3199	14,446	25	0.173
% lethals per brood					0.223	0.139	0.309	0.031			
Controls (concurrent)											
			17	68	4/1375	1/780	1/600	0/443	3204	6	0.187
			40	69	0/1520	2/1173	2/433	0/545	3675	4	0.109
Sum controls					4/2895	3/1953	3/1033	0/988	6879	10	0.145
% lethals per brood					0.138	0.153	0.097	0			

Table 42

"TRIS"

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
E	100	72	20	68	6/1301	4/660	2/592	1/646	3212	13	0.405
H	100	72	31	69	5/1446	9/989	1/560	1/694	3705	16	0.431
Sum experiments					11/2747	13/1649	3/1152	2/1340	6917	29	0.419
% lethals per brood					0.399	0.782	0.260	0.149			
Controls (concurrent)											
			17	68	4/1375	1/780	1/600	0/443	3204	6	0.187
			40	69	0/1520	2/1173	2/433	0/545	3675	4	0.109
Sum controls					4/2895	3/1953	3/1033	0/988	6879	10	0.145
% lethals per brood					0.138	0.153	0.097	0			

Table 43

"TRIS"

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	1000	72	16	63	26/2046	30/494	2/391	18/1061	4068	76	1.87
B	1000	72	14	64	27/1738	57/631	15/879	5/1157	4509	104	2.31
Sum experiments					53/3784	87/1125	17/1270	23/2218	8577	180	2.10
% lethals per brood					1.38	7.18	1.32	1.03			
Controls (concurrent)											
			4	63	3/2195	1/1097	1/753	2/825	4877	7	0.144
			1	64	3/1535	3/1124	1/808	1/572	4047	8	0.198
Sum controls					6/3730	4/2221	2/1561	3/1397	8924	15	0.168
% lethals per brood					0.161	0.180	0.128	0.214			

Tables 44 through 52

Summaries of experiments with reference mutagens. "Cluster" corrections were made only for controls and for the low mutagenicity runs (DBE and TMP).

Table 44

DBE: 1, 2 - DIBROMOETHANE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals	
A	5	72	8	54	2/2102	1/1335	2/1255	1/700	5398	6	0.111	
D	5	72	0	55	3/1491	1/662	0/1376	0/643	4176	4*	0.096	(0.334)
B	10	72	8	54	4/2330	2/1143	1/1235	1/528	5244	8**	0.153	(0.381)
E	10	72	0	55	4/1541	1/1056	1/1530	1/956	5090	7	0.138	
C	50	72	4	54	3/2504	2/1256	4/1568	0/709	6046	9***	0.149	(0.231)
F	50	72	9	55	0/2041	2/853	3/1113	2/911	4925	7	0.142	
Sum experiments					16/12009	9/6305	11/8077	5/4447	30,879	41	0.133	(0.220)
% lethals per brood					0.133	0.143	0.136	0.112				
Controls (concurrent)												
			3	54	0/1934	0/856	2/1284	0/606	4682	2	0.043	
			4	55	6/1480	1/435	0/853	1/409	3185	8	0.251	
Sum controls					6/3414	1/1291	2/2137	1/1015	7867	10	0.127	
% lethals per brood					0.175	0.077	0.094	0.098				

* Cluster of 10 deducted
** Cluster of 12 deducted
*** Cluster of 5 deducted
Numbers in parentheses are % lethals without removal of clusters

Table 45

ETHYLENIMINE (EI)

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals*	% lethals
D	50	72	20	47	21/137	0/0	8/310	8/534	1018	37	3.63
E	30	72	9	49	280/2302	79/940	15/1204	20/2362	7202	394	5.47
F	10	72	8	53	51/2494	**	12/1482	2/1036	5077	65	1.28
G	4	72	12	53	11/2340	**	12/1474	9/966	4812	32	0.665
H	2	72	12	53	10/2956	**	6/2135	4/1401	6512	20	0.307
Controls (concurrent)											
			48	47	2/1946	0/912	1/910	0/510	4281	3	0.070
			3	49	3/1310	2/1357	3/1116	1/700	4492	9	0.200
			12	53	3/1809	**	2/1447	0/912	4173	5	0.120

* Clusters not identifiable due to high mutation frequency.

** Broods 1 and 2 were combined.

Table 46

ETHYL METHANESULFOHATE (EMS)

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
M	2	72	0	43	14/1845	11/1337	7/947	2/604	4767	34	0.173
P	2	72	8	44	14/2416	7/1164	2/903	1/756	5461	24	0.439
Sum experiments					28/4261	18/2501	9/1850	3/1360	10,228	58	0.567
% lethals per brood					0.653	0.715	0.484	0.220			
Controls (concurrent)											
			0	43	3/2081	5/1708	7/1100	2/1052	5958	17*	0.285
			10	44	8/2622	3/1359	1/1236	1/767	5997	13	0.217
Sum controls					11/4703	8/3067	8/2336	3/1819	11,955	30	0.251
% lethals per brood					0.233	0.260	0.341	0.165			

* Cluster of 14 deducted

Table 47

ETHYL METHANESULFONATE (EMS)

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
A	4	48	1	33	8/1508	3/1024	2/1077	0/427	4049	13	0.321
N	4	72	0	43	20/1875	0/1411	4/975	1/580	4875	34	0.697
Q	4	72	8	44	10/2467	6/1265	2/808	0/733	5291	18	0.340
Sum experiments					38/5850	18/3700	8/2860	1/1740	14,215	65	0.457
% lethals per brood					0.645	0.484	0.279	0.057			
Controls (concurrent)											
			3	33	4/1295	4/1302	4/1355	0/728	4692	12*	0.255
			0	43	3/2081	5/1708	7/1100	2/1052	5958	17**	0.285
			10	44	8/2622	3/1359	1/1236	1/767	5997	13	0.217
Sum controls					15/5998	12/4369	12/3691	3/2547	16,647	42	0.252
% lethals per brood					0.249	0.274	0.324	0.118			

* Cluster of 12 deducted
** Cluster of 14 deducted

Table 48

ETHYL METHANESULFONATE (EMS)

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
E	10	48	0	36	18/2597	4/504	-	-	3123	22	0.704
O	10	72	0	43	33/2002	36/1488	14/986	3/701	5263	86	1.634
R	10	72	10	44	20/2366	9/1212	6/877	3/832	5325	38	0.714
Sum experiments					71/6965	49/3204	20/1863	6/1533	13,711	146	1.065
% lethals per brood					1.009	1.506	1.062	0.390			
Controls (concurrent)											
			1	36	6/1591	0/419	-	-	2016	6	0.298
			0	43	3/2081	5/1708	7/1100	2/1052	5958	17*	0.285
			10	44	8/2622	3/1359	1/1236	1/767	5997	13	0.217
Sum controls					17/6294	8/3486	8/2336	3/1819	13,971	36	0.258
% lethals per brood					0.269	0.229	0.341	0.165			

* Cluster of 14 deducted

Table 49

ETHYL METHANESULFONATE (EMS)

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
F	100	48	1	36	130/2014	27/222	-	-	2393	157	6.561
V	100	48	46	49	35/1222	36/1065	7/469	1/368	3203	79	2.466
W	100	48	2	49	59/1078	51/829	2/402	2/470	2893	114	3.941
Sum experiments					224/4314	114/2116	9/871	3/838	8489	350	4.123
% lethals per brood					4.936	5.112	1.023	0.357			
Controls (concurrent)											
			1	36	6/1591	0/419	-	-	2016	6	0.298
			3	49	3/1310	2/1357	3/1116	1/700	4492	9	0.200
Sum controls					9/2901	2/1776	3/1116	1/700	6508	15	0.230
% lethals per brood					0.309	0.112	0.268	0.143			

Table 50

ETHYL METHANESULFONATE (EMS)*

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
T	400	72	12	47	103/1508	19/537	5/921	4/876	3973	132	3.322
X	400	72	28	51	41/1670	27/1182	9/909	1/668	4507	78	1.731
U	100	5	20	49	12/1128	-	-	-	1140	12	1.053
Sum experiments T,X					144/3178	46/1719	14/1830	5/1544	8480	210	2.476
% lethals per brood					4.335	2.606	0.759	0.323			
Controls (concurrent)											
			48	47	2/1946	0/912	1/910	0/510	4281	3	0.070
			12	51	8/1895	1/1192	1/1076	1/541	4715	11	0.233
			3	49	3/1310	2/1357	3/1116	1/700	4492	9	0.200
Sum controls 47,51					10/3841	1/2104	2/1986	1/1051	8996	14	0.156
% lethals per brood					0.260	0.048	0.101	0.095			

* Experiments T and X: Flies exposed to vapor only
Experiment U: Flies exposed in regular feeding plates but with proboscis removed, to prevent feeding

Table 51

ETHYL METHANESULFONATE (EMS) SUMMARY OF COMPARABLE EXPERIMENTS

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests (1/1+) per brood 1	2	3	4	Total tests	No. lethals	% lethals
M,P	2	72	-	43,44	28/4261	18/2501	9/1850	3/1360	10,228	58	0.567
A,N,Q	4	48,72	-	33,43 44	38/5850	18/3700	8/2860	1/1740	14,215	65	0.457
E,O,R	10	48,72	-	36,43 44	71/6965	49/3204	20/1863	6/1533	13,711	146	1.065
F,V,W	100	48	-	36,49	224/4314	114/2116	9/871	3/838	8489	350	4.123
G	200	48	0	36	199/1439	83/531	-	-	2252	282	12.522
C	400	48	2	33	353/1586	267/1073	107/511	18/783	4698	620	13.197
% lethals per brood, exp. C					18.2	19.9	17.3	2.25			
Controls (concurrent)											
			-	43,44	11/4703	8/3067	8/2336	3/1819	11,955	30*	0.251
			-	33,43,44	15/5998	12/4369	12/3691	3/2547	16,647	42*	0.252
			-	36,43,44	17/6294	8/3486	8/2336	3/1819	13,971	36*	0.258
			-	36,49	9/2901	2/1776	3/1116	1/700	6508	15	0.230
			1	36	6/1591	0/419	-	-	2016	6	0.298
			3	33	4/1295	4/1302	4/1355	0/728	4692	12*	0.255
Sum controls					62/22782	34/14419	35/10834	10/7613	55,789	141*	0.253
% lethals per brood					0.271	0.235	0.322	0.131			

* Cluster of 12 deducted from run 33 and cluster of 14 from run 43

Table 52

TMP TRIMETHYL PHOSPHATE

Exp.	Conc. (ppm)	Time (hrs.)	% Mort.	Run	No. tests ($1/1^+$) per brood 1	2	3	4	Total tests	No. lethals	% lethals
B	1000	30,48	68	47	14/2201	5/992	6/1005	1/680	4904	26	0.530
C	300	24	0	67	2/1292	2/913	1/1010	7/995	4222	12	0.284
D	300	72	37	67	4/842	0/807	2/539	1/860	3055	7	0.229
E	100	24	0	67	1/1342	0/999	1/1248	0/992	4583	2*	0.044
F	100	72	27	67	0/1062	1/912	5/1028	2/917	3928	8**	0.204
Controls (concurrent)											
			48	47	2/1946	0/912	1/910	0/510	4281	3	0.070
			18	67	1/1214	1/1187	4/999	4/791	4201	10	0.238
Sum controls					3/3160	1/2099	5/1909	4/1301	8482	13	0.153

* Cluster of 9 deducted
** Cluster of 5 deducted

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